A Prototype for Blood Typing Based on Image Processing

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***Abstract*—This paper presents a new methodology for blood phenotyping based on the plate test and on image processing techniques to determine the occurrence of agglutination (between blood sample and reagent). A portable device for ABO-Rh blood typing and blood phenotying that automates all the analysis procedure, including mixture/centrifugation, reading and interpretation of results is presented. The system was tested with donor blood samples.**

Keywords - blood type determination; phenotyping; image processing techniques; prototype.

I. INTRODUCTION

The process of determination of blood types is essential before administering a blood transfusion, but in some cases, due to the risk of the individual´s life, it is necessary to quickly administer blood [1-3]. In these emergency situations, there is no time to determine the blood type and so the procedure is to administer the blood type O negative (universal donor) [1-3]. Nevertheless, due to some incompatibilities transfusion reactions may occur that could lead to the death of the patient [3].

Before performing a safe blood transfusion, certain compatibility tests, named pre-transfusion tests, must be undertaken, namely [4-6]:

1. Determine the A, B, AB, O (ABO system) and Rhesus (Rh) type of the patient;
2. Perform the reverse test: reverse grouping of ABO typing of the patient;
3. Perform the Rh (C, c, E and e) and Kell (K) phenotyping for detecting the presence of antigens in the blood of the patient. The search of other antigens can also be performed, but only in few situations [7];
4. Perform the screening of antibodies to detect the presence of significant antibodies. If antibody screening is positive, an identification of the antibody must be performed to allow the selection of the compatible blood;
5. Verify the results with previous data, if available;
6. Select the erythrocytes of the donor and perform the cross match.

The determination of ABO and Rh type can be performed in tube [8], plate [8], micro-plates [2], or gel centrifugation [9]. Due to the fast response time, the tube and the plate tests are those used in emergency situations. The micro-plates and gel centrifugation tests are more accurate than the tube or plate tests, but they require expensive and heavy devices.

This work aims to develop a prototype able to automatically perform the pre-transfusion tests necessary for a safe blood transfusion. This device is based on a previous version of the equipment developed by the authors [10-13] and it automates the reading, centrifugation and interpretation of results of all the pre-transfusion tests previously mentioned. The prototype presented is able to determine ABO and Rh blood typing and blood phenotyping.

It is important to mention that currently these tests are performed manually or at least semi-automatically, which may occur in errors in the testing procedure, the reading and interpretation of the results, being sometimes fatal to the patient [14, 15].

Apart from the present section, the paper is organized with more four sections. In Section II, the phenotyping determination is described, presenting the image processing techniques applied. The prototype developed is presented in Section III and in Section IV the results of the phenotyping ABO and Rh tests with the prototype developed are discussed. Finally, Section VI presents the conclusions and the directions for future work.

II. PHENOTYPING DETERMINATION

A methodology was developed regarding the pre-transfusion tests for determining the ABO and Rh type, based on the plate test and image processing techniques [13, 16-18].

This work is focused on the phenotyping tests Rh (C, c, E and e) and Kell (K), based also on the plate test [8]. The phenotyping determination is obtained through image processing techniques applied to the target image. The plate test method was employed due to its reliability and fast response time suitable in emergency situations.

The procedure used in the phenotyping is described in [19], where it can be observed that it requires centrifugation to separate the blood from the plasma and red blood cells, which is a costly and complex process.

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The proposed system is able to perform the centrifugation in a simple approach. After separating the blood and obtaining the plasma, six slides are considered, one for each of the phenotypes tested. In each slide, it was added a drop of the respective reagent TransClone Anti-RH1 (D), TransClone Anti-RH2 (C), Transclone Anti-RH3 (E), TransClone Anti-RH4 (c), TransClone Anti-RH5 (e) and TransClone Anti-Kel1 (K) [19], and a drop of blood of the patient. The mixture was manually performed using a glass rod in each slide. An image of the sample (mixed blood/reagent) was acquired and sent to the computer for further processing. The presence (or absence) of agglutination in each sample was determined through image processing techniques. The classification algorithm used in the analysis is implemented to work with blood samples after its centrifugation. The camera used to capture the images was a Sony Cyber-shot DSC-S750 of 7.2 Megapixel with 3X Optical Zoom (35-105 mm eq.) and 5 point auto focus [20].

To process the images, in order to determine the blood phenotyping as well as ABO-Rh, an algorithm was developed using IMAQ Vision software from National Instruments [21]. The algorithm developed is presented in Figure 1, as a flowchart [21-23]. The image captured is segmented, the localization of each of the wells is obtained and then the mixture reagents/blood is quantified, among others, with the standard deviation value.

In Figure 1 are presented the image processing techniques applied: image selection and storage, extraction of the color planes of the image, application of thresholds and fill holes, removal of small and border objects, and analysis. The original image is retrieved and luminance planes are removed, pattern and geometric matching is used to identify the imagine reference and containers position; finally the quantify function is employed determining the blood type in analysis.

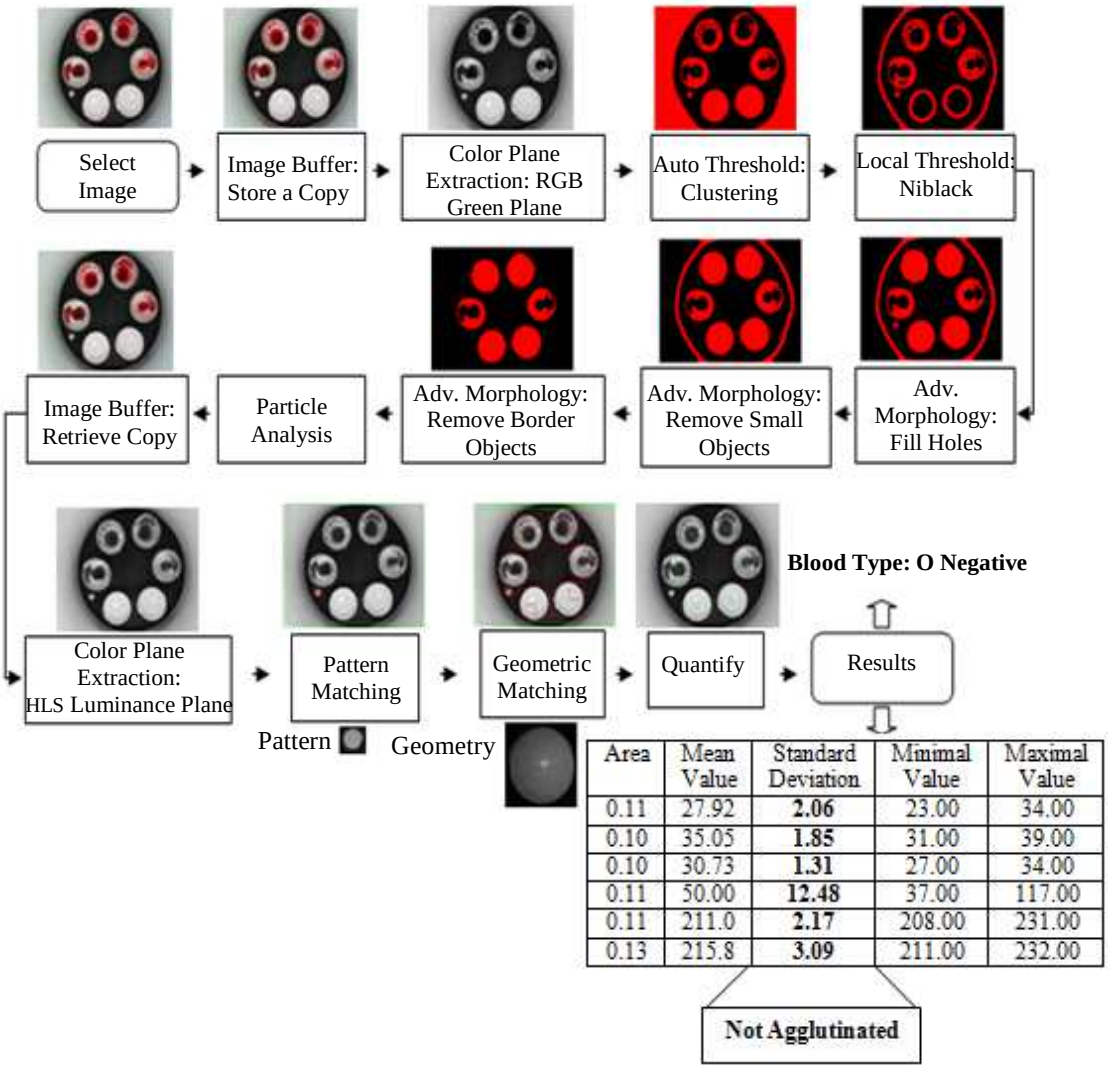

Figure 1. Flowchart of the algorithm developed for the prototype.

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III. AUTOMATIC AND PORTABLE SYSTEM DEVELOPED

The proposed system was developed to automatically determine the human blood type of a patient (phenotyping and ABO-Rh). An important requisite of the developed system was its portability (small and easy to transport to any place) in order to be used in emergency vehicles and other services. Thus, the analysis can be performed outside the clinical analysis laboratory and the results can be sent to the hospital through Wi-Fi or GSM.

The system developed has 30 cm of height and 10,5 cm of diameter (Figure 2). It has an AC motor that allows the mixture and the centrifugation. The motor rotational speed is adjustable depending if the test performed is a mixture (low speed) or a centrifugation (high speed). The motor can spin the plate test with the six containers and mixture the blood and reagent, or spin the plate and centrifuge the blood for obtaining the plasma. When the test is performed the motor is stopped, the LEDs switch on and then an image with all the necessary data is captured. The image is processed and the result of the test is given. At the end the LEDs switch off. The whole procedure is illustrated in the flowchart of Figure 3.

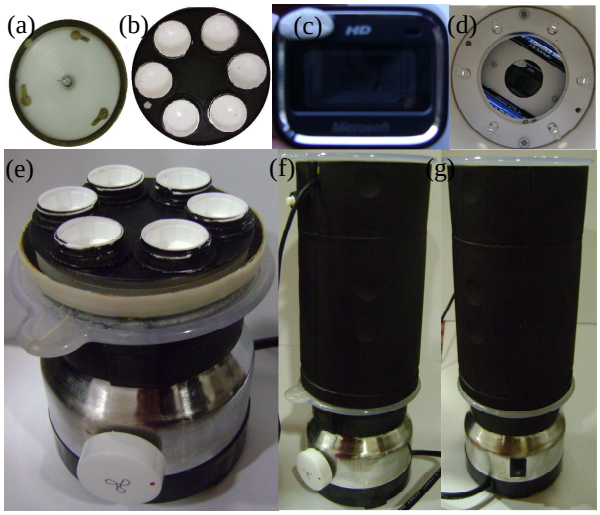

Figure 2. Portable System developed: (a) base of the system where piece with containers fits into the system, (b) piece with containers for mixture of blood and reagents, (c) web camera used to capture the image, (d) LEDs around and web camera in the centre, (e) base of the system that promote the mixture, (f) final system with button to change velocity, (g) final system with switch to switch on/off the system.

The user friendly application where all the image processing techniques are included as well as the classification algorithm was developed in C# Language and XAML (Figure 4).

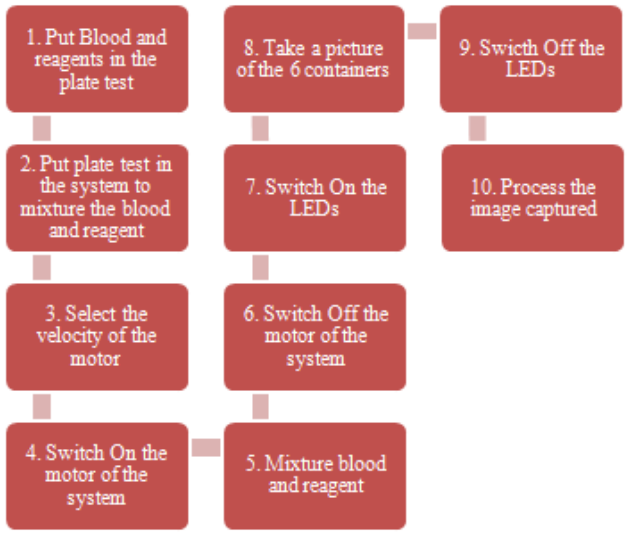

Figure 3. Flowchart of the system developed.

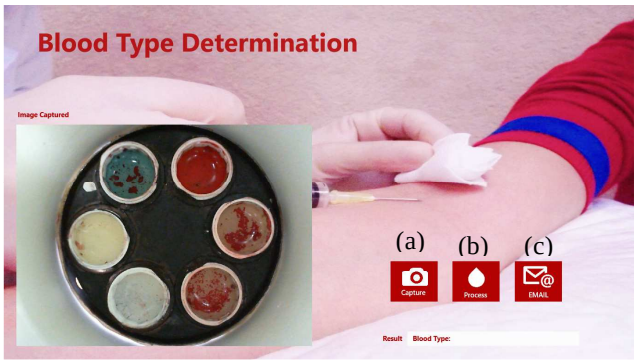

Figure 4. Application Developed. (a) Button to capture the image. (b) Button to process the image captured. (c) Button to send the results message.

This application contains a button to capture the original image with the six containers, Figure 4 (a), a button to process the original image and give the blood type result, Figure 4 (b), and finally a button to send a message to the laboratory with the blood type result, Figure 4 (c).

IV. RESULTS OBTAINED WITH THE DEVELOPED SYSTEM

This section presents the results of phenotyping tests (A) and the results of ABO and Rh tests (B), obtained with the prototype developed. The blood samples tested were from donors from the Portuguese Blood Institute (IPST).

A. Results of phenotyping tests

The methodology presented in this paper was tested and validated using several blood phenotypes from donors. This subsection presents the obtained results when applying the image processing techniques developed in images captured after the procedure of plate test (Section II). Figure 5 shows

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six original images corresponding to the mixture of blood and the respective reagent, TransClone Anti-RH1 (D), TransClone Anti-RH2 (C), Transclone Anti-RH3 (E), TransClone Anti-RH4 (c), TransClone Anti-RH5 (e) and TransClone Anti-Kel1 (K), captured by the camera. Figure 6 shows the images obtained after the application of image processing techniques and in Table I are summarized the quantified results.

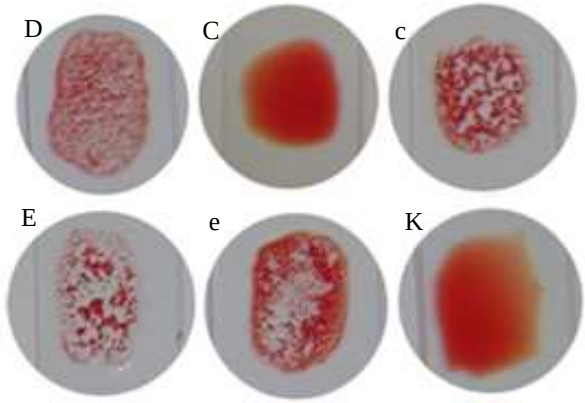

Figure 5. Images captured of blood/reagent mix for determination of Rh and Kell phenotype.

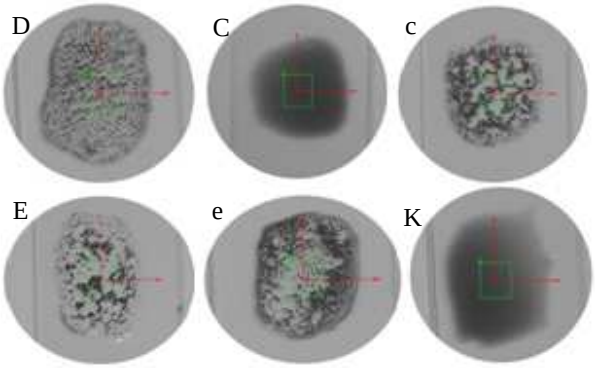

Figure 6. Results of the application of the developed algorithm through the images of Figure 4.

TABLE I. RESULTS OF THE DEVELOPED ALGORITHM THROUGH THE IMAGES OF FIGURE 5

Figure 6	Area	Mean Value	Standard Variation	Minimal Value	Maximal Value
D	3,19	118,36	**29,09**	21,00	181,00
C	2,08	59,24	3,08	51,00	71,00
c	1,98	118,69	**39,32**	20,00	184,00
E	2,67	125,20	**44,91**	15,00	195,00
e	3,75	131,88	**29,30**	25,00	195,00
K	3,18	67,31	5,70	55,00	86,00

Analyzing Figure 5, one can see that agglutination occurred in images corresponding to phenotype test D, c, E and e (Standard Variation values highlighted in bold), and agglutination does not occur in the remaining images C and K. It can be observed in Table I, that when agglutination occurs, images D, c, E and e of Figure 6, present a standard deviation value higher than 16 (threshold established in previous studies [13] to detect the occurrence of agglutination). The standard deviation value is the threshold for determining the occurrence or non occurrence of agglutination. Thus, standard deviation value above 16 indicates the occurrence of agglutination, while standard deviation values below 16, mean the non occurrence of agglutination in the analyzed image. Thus, given that agglutination occurred for phenotypes D, c, E and e, and agglutination was not observed for other phenotypes, C and K, the phenotype under analysis is then cEeK-D (confirmed by the analysis performed by the IPST).

B. Results of ABO and Rh tests

The proposed system allows also ABO and RH blood typing [12, 13]. In Figure 7, it is presented the original image of the containers captured after the mixture of blood sample and the specific reagents. Figure 8 presents the final image after the application of the developed image processing algorithm. TABLE II presents the occurrence of agglutination in each container and the correspondent ABO-Rh blood type. The agglutination occurred in Figure 8 with the reagents Anti-B, Anti-AB and Anti-D and not in Figure 8 with the reagent Anti-A, meaning that in this blood sample are present the antigens of type B and D and not the antigens of type A. The blood type is then B positive.

The determination of the occurrence of agglutination in the ABO-Rh blood test is also performed through the value of standard deviation.

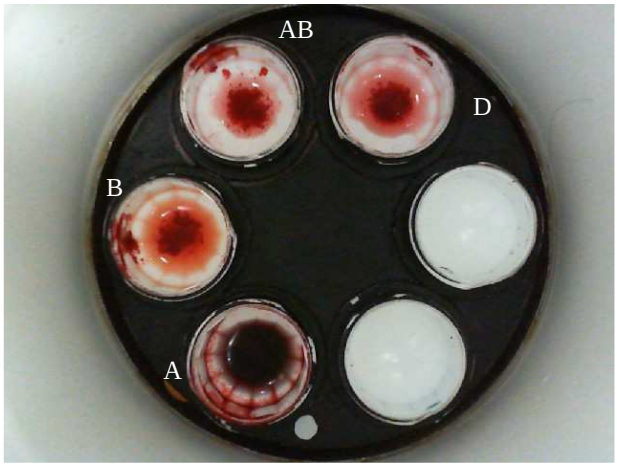

Figure 7. Image captured of blood/reagent mix for determination of ABO and Rh.

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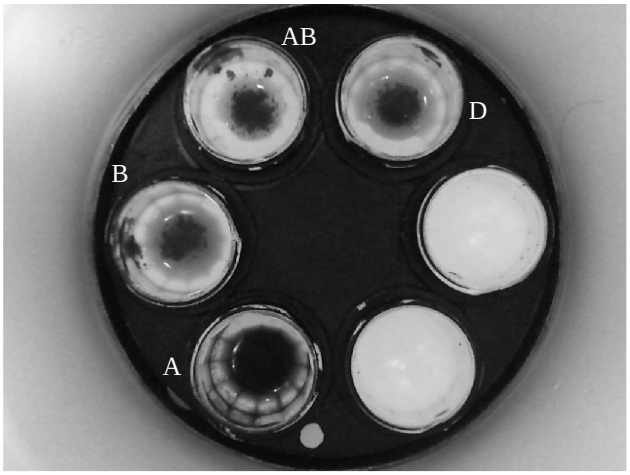

Figure 8. Results of the application of the developed algorithm through the images of Figure 6

TABLE II. RESULTS OF THE DEVELOPED ALGORITHM THROUGH THE IMAGES OF FIGURE 6

Figure 7	Occurrence of Agglutination	Blood Type
A	No	B
B	Yes	
AB	Yes	
D(Rh)	Yes	Positive

By observing TABLE II, it is possible to conclude that the blood type resultant is B Positive, as agglutination occurred with the reagents anti-B, anti-AB and anti-D.

V. CONCLUSION AND FUTURE WORK

This work allows enforcing the validation of the use of the plate test and image processing techniques as a feasible methodology to blood typing. As in [13, 16-18], the plate test has proven to be adequate to detect the occurrence of agglutination, even if it is weak, and the image processing techniques developed allow a correct automatic detection of agglutination, permitting ABO-RH blood typing as well as the phenotypes of the patient.

Once the system obtains results in a short time, about 5 minutes in total, at a low cost and being portable, it is characterized by an innovative solution with commercial added value. The system can be either used for dual operation, mixture (low speed) and centrifuging (high speed), depending of the motor velocity selected. Due to its portability and fast response time it can be used in ambulances or other vehicles for emergency situations.In future, it is intended to include closed loop control to the illumination and the rotational speed of the motor as well as, introduce neural networks to improve the classifications of the developed system. With the neural networks it will be possible to classify blood types as well as pre-transfusion tests more efficiently, quickly and safely.

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